

## Dissecting spermathecae to determine insemination status V 2

### **Background:**

It is often necessary to determine whether females in a colony are inseminated. Failure to mate can result in diminished colony production due to disease, or inadequate mating stimulus. The quickest and easiest method is to check the rate of insemination in females, for which we suggest dissection of the spermathecae and observation for presence of sperm.

### **Solution:**

You will need a typical stereo and compound scope.

- 1) Anesthetize the adult females.
- 2) Place a drop of PBS on a clean slide.
- 3) Gently grasp female by the thorax with forceps, and place ventral side up in the PBS on the slide under the stereoscope.
- 4) While viewing the specimen under the stereoscope, take a fine tip needle or forceps and gently remove the terminalia of the female by grasping them and pulling away slowly.
- 5) Locate the spermathecae. It should appear as a dark “golf-ball” like object that may or may not be surrounded by accessory tissues. Dispose of the mosquito and remaining tissues including the terminalia.
- 6) Gently lower a cover slip onto the spermathecae using a needle (to avoid rupturing the spermathecae). On the underside of the slide circle the area surrounding the spermathecae using a permanent marker. The marking will make it much simpler to locate the organ.
- 7) Under 100X magnification, look for the telltale wriggling of the long thread-like sperm. They will appear as fine concentric threads within the spermathecae. If rupturing has occurred, scan the surrounding field for the sperm.

